

SEQUENCE LISTING

<110> Chr. Hansen A/S  
Maarten van den Brink, Johannes  
Harboe, Marianne K  
Petersen, Steen Guldager  
Rahbek-Nielsen, Henrik

<120> IMPROVED METHOD OF PRODUCING AN ASPARTIC PROTEASE POLYPEPTIDE IN  
A RECOMBINANT HOST ORGANISM

<130> P1031US00

<150> PA 2002 0092  
<151> 2002-06-17

<160> 8

<170> PatentIn version 3.3

<210> 1  
<211> 323  
<212> PRT  
<213> Bos taurus

<400> 1

Gly Glu Val Ala Ser Val Pro Leu Thr Asn Tyr Leu Asp Ser Gln Tyr  
1 5 10 15

Phe Gly Lys Ile Tyr Leu Gly Thr Pro Pro Gln Glu Phe Thr Val Leu  
20 25 30

Phe Asp Thr Gly Ser Ser Asp Phe Trp Val Pro Ser Ile Tyr Cys Lys  
35 40 45

Ser Asn Ala Cys Lys Asn His Gln Arg Phe Asp Pro Arg Lys Ser Ser  
50 55 60

Thr Phe Gln Asn Leu Gly Lys Pro Leu Ser Ile His Tyr Gly Thr Gly  
65 70 75 80

Ser Met Gln Gly Ile Leu Gly Tyr Asp Thr Val Thr Val Ser Asn Ile  
85 90 95

Val Asp Ile Gln Gln Thr Val Gly Leu Ser Thr Gln Glu Pro Gly Asp  
100 105 110

Val Phe Thr Tyr Ala Glu Phe Asp Gly Ile Leu Gly Met Ala Tyr Pro  
115 120 125

Ser Leu Ala Ser Glu Tyr Ser Ile Pro Val Phe Asp Asn Met Met Asn  
130 135 140

Arg His Leu Val Ala Gln Asp Leu Phe Ser Val Tyr Met Asp Arg Asn  
145 150 155 160

Gly Gln Glu Ser Met Leu Thr Leu Gly Ala Ile Asp Pro Ser Tyr Tyr  
165 170 175

Thr Gly Ser Leu His Trp Val Pro Val Thr Val Gln Gln Tyr Trp Gln  
180 185 190

Phe Thr Val Asp Ser Val Thr Ile Ser Gly Val Val Val Ala Cys Glu  
195 200 205

Gly Gly Cys Gln Ala Ile Leu Asp Thr Gly Thr Ser Lys Leu Val Gly  
210 215 220

Pro Ser Ser Asp Ile Leu Asn Ile Gln Gln Ala Ile Gly Ala Thr Gln  
225 230 235 240

Asn Gln Tyr Gly Glu Phe Asp Ile Asp Cys Asp Asn Leu Ser Tyr Met  
245 250 255

Pro Thr Val Val Phe Glu Ile Asn Gly Lys Met Tyr Pro Leu Thr Pro  
260 265 270

Ser Ala Tyr Thr Ser Gln Asp Gln Gly Phe Cys Thr Ser Gly Phe Gln  
275 280 285

Ser Glu Asn His Ser Gln Lys Trp Ile Leu Gly Asp Val Phe Ile Arg  
290 295 300

Glu Tyr Tyr Ser Val Phe Asp Arg Ala Asn Asn Leu Val Gly Leu Ala  
305 310 315 320

Lys Ala Ile

<210> 2

<211> 1142

<212> DNA

<213> artificial

<220>

<223> DNA fragment comprising a DNA fragment of 1138 bp designed to comprise a N-H-T glycosylation site and unique SalI and XhoI sites for cloning purposes (modB-XS).

<400> 2

cggtcgaccg	ctacggtgac	tgacacacctgg	cgtgccgaga	tcactcgcat	ccccctctac	60
aaggcaagt	ctctgcgtaa	ggctctcaag	gagcacggtc	tgctcgagga	tttcctgcag	120
aagcagcagt	acggcatcaag	ctctaagtac	agcggttcg	gcgaggtggc	cagcgtgcct	180
ctcactaact	acctggacag	ccagtgacttc	ggtaagatct	accttggcac	tccccctcag	240
gagttcaccg	ttctgttcga	tactggttcc	agcgacttct	gggttccctc	catctactgt	300
aagagcaacg	cttgcaagaa	ccaccagcgc	ttcgatccctc	gcaagtccag	cacccctccag	360
aacccctggca	agcccccatttc	catccactac	ggtactggca	gcatgcaggg	tatccttggc	420
tacgacaccg	ttaccgtgtc	caacatcgtc	gatattcagc	agaccgtggg	tctgagcacc	480
caggagcctg	gcgatgtctt	cacttacgcc	gagttcgatg	gtatcctcg	catggcttac	540
ccctccctgg	cctctgagta	ctctatccct	gtgtcgaca	acatgatgaa	ccggccaccc	600
gtcgctcagg	atctgttcag	cgtgtacatg	gaccgtaacg	gtcaggagtc	catgcttact	660
ctgggcgcca	tcgatccctc	ttactacacc	ggttccctcc	actgggttcc	tgtgaccgtc	720
cagcagtaact	ggcagttcac	cgtggacagc	gtcactatct	ccggcgtgg	tgtggcttgc	780
gagggtggct	gtcaggccat	ccttgatact	ggtaccagca	agctcgtcgg	cccccctccagc	840
gacatcctga	acatccagca	ggctatcggt	gccacccaga	accagtaacgg	cgagttcgat	900
atcgactgcg	ataacccatttc	ttacatgcct	actgtggttt	tcgagatcaa	cggtaagatg	960
tacccccccta	ctccttctgc	ttacacttcc	caggatcagg	gcttctgtac	ctctggtttc	1020
cagtcgtgaga	accacagcca	gaagtggatc	cttggcgatg	tcttcatccg	cgagtactac	1080
tccgttccg	accgtgccaa	caacctggtg	ggtctcgcta	aggccatctg	atcctctaga	1140
gt						1142

<210> 3  
 <211> 408  
 <212> DNA  
 <213> artificial

<220>  
 <223> an approximately 410 bp SalI-SphII I fragment made using  
 synthetic oligonucleotides (SEQ ID XXX-1)

<400> 3	cggtcgaccg	ctacggtgac	tgacacacctgg	cgtgccgaga	tcactcgcat	ccccctctac	60
aaggcaagt	ctctgcgtaa	ggctctcaag	gagcacggtc	tgctcgagga	tttcctgcag	120	
aagcagcagt	acggcatcaag	ctctaagtac	agcggttcg	gcgaggtggc	cagcgtgcct	180	
ctcactaact	acctggacag	ccagtgacttc	ggtaagatct	accttggcac	tccccctcag	240	
gagttcaccg	ttctgttcga	tactggttcc	agcgacttct	gggttccctc	catctactgt	300	

aagagcaacg cttgcaagaa ccaccagcgc ttcgatcctc gcaagtccag caccttccag 360  
aaccttggca agccccttc catccactac ggtactggca gcatgcag 408

<210> 4  
<211> 233  
<212> DNA  
<213> artificial

<220>  
<223> an approximately 220 bp SphI-BsrGI fragment made using synthetic oligonucleotides (SEQ ID XXX-2)

<400> 4  
gcagcatgca gggtatcctt ggctacgaca ccgttaccgt gtccaaacatc gtcgatattc 60  
agcagaccgt gggcttgagc acccaggagc ctggcgatgt cttcaattac gccgagttcg 120  
atggtatcct cggcatggct taccctccc tggcctctga gtactctatc cctgtgttcg 180  
acaacatgat gaaccgccac ctctgtcgctc aggatctgtt cagcgtgtac atg 233

<210> 5  
<211> 200  
<212> DNA  
<213> Artificial

<220>  
<223> an approximately 190 bp BsrGI-KpnI fragment made using synthetic oligonucleotides (SEQ ID XXX-3)

<400> 5  
gcgtgtacat ggaccgtaac ggtcaggagt ccatgcttac tctggcgcc atcgatccct 60  
cttactacac cggttccctc cactgggttc ctgtgaccgt ccagcagttac tggcagttca 120  
ccgtggacag cgtcaactatc tccggcgtgg ttgtggcttg cgagggtggc tgtcaggcca 180  
tccttgatac tggtaccagc 200

<210> 6  
<211> 334  
<212> DNA  
<213> artificial

<220>  
<223> an approximately 320 bp KpnI-XbaI fragment made using synthetic oligonucleotides (SEQ ID XXX-4)

<400> 6  
ctggtaccag caagctcgtc ggcccctcca gcgacatcct gaacatccag caggctatcg 60  
gtgccaccca gaaccagtac ggcgagttcg atatcgactg cgataacctt tcttacatgc 120  
ctactgttgt tttcgagatc aacggtaaga tgtacccct tactccttct gcttacactt 180  
cccaggatca gggcttctgt acctctgggt tccagtctga gaaccacagc cagaagtgg 240

tccttggcga tgtcttcatc cgcgagtaact actccgtctt cgaccgtgcc aacaacctgg 300

tgggtctcgc taagggcatac tgatcctcta gagt 334

<210> 7

<211> 334

<212> DNA

<213> artificial

<220>

<223> a modified KpnI-XbaI fragment designed for construction of the modBM gene (SEQ ID XXX-5).

<400> 7

ctggtaaccag caagctcgta ggccccctcca gcgcacatcctt gaacatccag caggctatcg 60

gtgccaccca gaaccagttac ggcgagttcg atatcgactg cgataacctt tcttacatgc 120

ctactgtggt tttcgagatc aacggtaaga tgtacccctt tactccttctt gcttacactt 180

cccaggatca gggcttctgt acctctgggt tccagtctga gaaccacacc cagaagtgg 240

tccttggcga tgtcttcatc cgcgagtaact actccgtctt cgaccgtgcc aacaacctgg 300

tgggtctcgc taagggcatac tgatcctcta gagt 334

<210> 8

<211> 66

<212> DNA

<213> artificial

<220>

<223> synthetic polylinker (SalI-SphI-BsrGI-KpnI-XbaI) (SEQ ID XXX-6)

<400> 8

ggccaggcgc gccttccatg gaagaatgcg gccgctaaac catcgatggc tcgagttggc 60

gcggca 66